

1994. ABSTRACT No. 93. Page 17 in SETAC 15th Annual Meeting  
(October 30-November 3, 1994, Denver, Colorado)

**93 A Novel Extraction Method for the Analysis of Cholecalciferol-Treated Oat Baits.** R.E. Mauldin, M.J. Goodall, and D.A. Goldade, USDA/APHIS, Denver Wildlife Research Center, Denver, CO. Cholecalciferol (Vitamin D<sub>3</sub>) is used as a rodenticide and is frequently applied in a grain bait matrix. Many analytical methods for cholecalciferol determination require an additional saponification step resulting in isomerization of cholecalciferol to pre-cholecalciferol. While equivalent biologically, pre-cholecalciferol is difficult to quantify accurately. An extraction procedure was developed utilizing a solvent mixture of 97% n-hexane/3% isopropanol which also served as the mobile phase for a subsequent liquid chromatographic analysis. One gram samples of Quintox® (Motomco Ltd, Madison, WI)-treated steamed, rolled oats fortified to 0.05% or 0.75% cholecalciferol (w/w) were extracted by rapid shaking. Final concentrations of the extracts were 10 and 150 µg/mL, respectively. Analyses were performed on a liquid chromatograph, using a silica stationary phase analytical column (3 µm, 4.6-mm x 25 cm), with a flow rate of 2.0 mL/min and a wavelength of UV 265 nm. A linear ( $r^2 = 0.9993$ ), directly proportional response in cholecalciferol standards ranging in concentration from 10 to 150 µg/mL was found. No matrix interferences were observed. Method limit of detection was equal to 0.0016% cholecalciferol. Recoveries of 99 and 98% were observed in seven replicates of the 0.05 and 0.75% fortified baits, respectively. Formation of pre-cholecalciferol was negligible.